Remarks

Amendments to the Specification

Applicants have amended the specification to remove hyperlinks in compliance with, and thus obviating, the Examiner's objection. No new matter has been added.

Rejections under 35 U.S.C. § 112, ¶ 2 Are In Error And/Or Have Been Obviated By Amendment And Should Be Withdrawn

The Examiner rejects claims 6 and 9 for recitation of the limitation "differentially displayed," and rejects claim 7 for recitation of "differentially expressed," both on grounds that the phrases are unclear. Office action at items 5, 6, 7, and 9.

Applicants respectfully disagree.

A differentially displayed protein is a protein that is differentially detectable by mass spectrometry as between two samples: in a display of the mass spectra of the respective samples, ions of the protein are thus differentially displayed. At the limit, the difference in detectability can be absolute: the protein's ions are displayed in one but not the other of the mass spectra. In many cases, however, the difference is less than absolute, with the mass spectral difference being one of ion abundance, or apparent m/z, rather than an absolute presence or absence. The differentially displayed protein is typically, but not invariably, differentially present within the two samples, as may occur when the protein is

differentially expressed within cells from which the respective samples are derived.

See, e.g., specification at 109, lines 11 - 18, specification at 105, lines 1 - 12, and original claim 9.

Applicants submit that the terms would readily and unambiguously have been so understood by the skilled artisan.

However, solely to expedite prosecution, applicants have clarified claims 6 and 7 by amendment more particularly to point out and distinctly change their invention. Applicants respectfully submit that the language of claim 9, which has not been amended, is clear and definite in light of the amendments to claims 6 and 7. No new matter has been added.

The Examiner further rejects claim 7 for lack of antecedent basis for recitation of the claim element "protein biochip." Applicants have amended the phrase to recite instead "affinity capture probe" and respectfully submit that the amendment has obviated the rejection.

Rejections Under 35 U.S.C. 103 Are In Error And/Or Have Been Obviated And Should Be Withdrawn

In a first rejection under 35 U.S.C. § 103, the Examiner rejects claims 6 and 8 - 14 as having been obvious over Hutchens *et al.* (WO 98/59362, "Hutchens") in view of Dongre et al. (TIBTECH vol. 15, October 1997, "Dongre"). Claim 7 is free of this rejection.

Applicants respectfully disagree.

Applicants' claimed methods facilitate the identification of a protein that is differentially displayed in the mass spectra of two samples by correlating the characteristics of a differentially displayed protein fragment with the characteristics of a differentially displayed protein. This back-correlation of fragment properties to protein properties enhances the reliability with which a database call of putative identity is made, particularly when the differentially displayed protein is present in complex admixture with other proteins in one or both of the samples.

When one or both of the protein and fragment spectra is obtained by first capturing the respective protein or fragment on a surface-enhanced laser desorption ionization (SELDI) probe, the methods of the present invention provide additional physicochemical characteristics of protein and fragment that can be used to increase the reliability of protein identification. With on-probe digestion and a tandem mass spectrometer having a SELDI probe interface, the methods of the present invention provide particularly rapid and efficient identification, providing a substantial improvement over methods in the art.

Even were the Examiner's combination of the two references adequately suggested or motivated in the prior art, which applicants respectfully traverse,[2] the reference

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[2] It is by now well settled that the Examiner must show a motivation to combine references to establish a *prima facie* case of obviousness. *In re Rouffet*, 47 USPQ2d 1453 (Fed. Cir. 1998) ("To prevent the use of hindsight

combination neither describes nor suggests applicants' invention as a whole.

Of the four successive steps in the method of applicants' claim 6 as now pending, Hutchens may be said to disclose step (a), detecting at least one protein that is differentially displayed in the mass spectra of two samples. Separately, Hutchens also discloses step (b), cleaving proteins in the two samples and then detecting protein cleavage products that are differentially displayed in the mass spectra of the two cleaved samples.

As the Examiner notes, Hutchens does not disclose applicants' step (c), determining the identity of a differentially displayed protein cleavage product with a tandem mass spectrometer. Not mentioned by the Examiner, Hutchens also critically fails to disclose applicants' step (d),

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based on the invention to defeat patentability of the invention, this court requires the examiner to show a motivation to combine the references that creates the case of obviousness."); *In re Dembiczak*, 50 USPQ2d 1614 (Fed. Cir. 1999); *WMS Gaming Inc. v. International Game Technology*, 51 USPQ2d 1385 (Fed. Cir. 1999); *Brown & Williamson Tobacco Corp. v. Philip Morris Inc.*, 56 USPQ2d 1456 (Fed. Cir. 2000); *In re Lee*, 61 USPQ2d 1430 (Fed. Cir. 2002).

"The factual inquiry whether to combine references must be thorough and searching. It must be based on objective evidence of record. The need for specificity pervades this authority." *In re Lee*, 61 USPQ2d at 1433 (internal quotations and citations omitted). "[P]articular findings must be made as to the reason the skilled artisan, with no knowledge of the claimed invention, would have selected these components for combination in the manner claimed." *In re Kotzab*, 217 F.3d 1365, 1371, 55 USPQ2d 1313, 1317 (Fed. Cir. 2000).

Absent such thorough, searching, objective, specific and particularized findings, there can be no *prima facie* case, *In re Lee*, 61 USPQ2d 1430, 1433 (Fed. Cir. 2002); absent a *prima facie* case, an applicant is entitled, without more, to his patent, *In re Glaug*, 62 USPQ2d 1151 (Fed. Cir. 2002).

correlating the identity of the protein cleavage product with a differentially displayed protein as detected in step (a).

Dongre is a review of tandem mass spectrometry techniques for the rapid identification of proteins, and discloses a typical tandem MS (MS/MS) identification of a protein of interest, in which the product (*i.e.*, fragment) ion scan of a selected protein cleavage product is used for database query. At most, Dongre could arguably be said to disclose a portion of applicants' step (c): determining the identity of a protein cleavage product by MS/MS analysis. Dongre does not disclose the entirety of applicants' step (c), however, since the peptide isolated by MS1 for fragmentation and subsequent mass spectral analysis in MS2 (see Dongre FIG. 1) is never said to have been chosen based upon its differential display as between two samples.

It is beyond debate that Dongre does not disclose applicants' step (a), step (b), or step (d).

Even if there were a suggestion or motivation in the prior art to combine the two references that would have been sufficient to establish a *prima facie* case of obviousness, the combination fails to teach, disclose, suggest, motivate, or render obvious applicants' invention as a whole. The rejection is in error and should thus be withdrawn.

In a second rejection, the Examiner rejects claims 6 and 8 - 14 under 35 U.S.C. § 103 as having been obvious over

Liebler et al. (U.S. Pat. No. 6,379,970) in view of Dongre. Claim 7 is free of this rejection.

Applicants disagree.

Liebler does not teach applicants' step (a): first detecting at least one protein that is differentially displayed in the mass spectra of the two samples. Indeed, Liebler's invention is intended to obviate that step:

> "[t]he present invention is based, in part, on detecting the differential expression of the same protein in two [s]amples . . . by analysis of peptide fragments from each sample. To that end, the method of the present invention includes digesting the protein into samples to a mixture of peptides and then comparing the abundances of specific peptides."

Liebler col. 3, lines 31 - 37. *See also* Liebler FIGS. 1 and 2, and Liebler claim 1, the first step of which claimed method is "digesting the protein in a plurality of biological samples to produce peptides in each sample."

Liebler does not teach applicants' step (b): cleaving proteins in the two samples into protein cleavage products ***and with a mass spectrometer detecting*** protein cleavage ***products that are differentially displayed*** in the mass spectra of the two cleaved samples. Liebler is quite clear that, following digestion, the peptides are labeled and then separated and detected by a method selected from the group consisting of "2D gel electrophoresis, capillary electrophoresis, isoelectric focusing and high-performance liquid chromatography (HPLC),"

particularly reverse phase HPLC. Liebler col. 6, lines 20 - 30 ("Peptide Separation").

> In a preferred embodiment, the samples containing the labeled peptides are combined prior to separation. In this embodiment, a single analytical step, e.g., a single HPLC separation, produces the data necessary to identify the differentially expressed proteins in the original samples.

Liebler col. 6, lines 50 - 54. Only after such identification is made does Liebler propose the use of mass spectrometry.

As the Examiner acknowledges, Liebler does not teach applicants' step (c), determining the identity of at least one differentially displayed protein cleavage product ***with a tandem mass spectrometer***: "Liebler et al does not particularly point out using tandem mass spectrometry as recited in claim 6," Office Action, p. 8.

And there should be no argument that Liebler does not disclose applicants' step (d).

All told, Liebler discloses ***not one*** of the steps of applicants' claimed methods.

For the reasons advanced above, such global defects are hardly remedied by Dongre.

Even if there were suggestion or motivation in the prior art to combine the two references that would have been sufficient to establish a *prima facie* case of obviousness, the combination fails to teach, disclose, suggest, motivate, or

render obvious applicants' invention as a whole. The rejection is in error and should thus be withdrawn.

In a final rejection under section 103, the Examiner rejects claim 7 as unpatentable over Hutchens in view of Dongre and further in view of Little et al. (U.S. Pat. No. 6,322,970, "Little").

The Examiner offers Little for purpose of "teach[ing] using a microchip to isolate a polypeptide as well as a means to manipulate the isolate target polypeptide prior to mass spectrometry," Office action p. 9, item 13. Given the inadequacy of the Hutchens and Dongre combination to render obvious the method of applicants' broader claim 6, the addition of Little does little to remedy the added deficiency in rendering obvious an element additional to claim 7

Furthermore, as amended herein pursuant to the Examiner's concerns under section 112, second paragraph, claim 7 no longer recites "biochip", instead reciting "capturing proteins from the samples on affinity capture probes."

> **"Affinity capture probe"** refers to a probe that binds analyte through an interaction that is sufficient to permit the probe to extract and concentrate the analyte from an inhomogeneous mixture. Concentration to purity is not required. The binding interaction is typically mediated by adsorption of analyte to an adsorption surface of the probe. Affinity capture probes are often colloquially referred to as **"protein biochips"**, which phrase is thus used herein synonymously with "affinity capture probe". The term **"ProteinChip® Array"** refers to affinity capture probes that are

> commercially available from Ciphergen Biosystems, Inc., Fremont, California, for use in the present invention. Affinity capture probes can have chromatographic adsorption surfaces or biomolecule affinity surfaces, as hereinafter defined.

Specification p. 38, line 25 - p. 39, line 8.

The Examiner has pointed to no disclosure within Little that teaches an affinity capture probe, as so defined.[3]

Thus, even if there were a suggestion or motivation in the prior art to combine the three references that would have been sufficient to establish a *prima facie* case of obviousness, the combination fails to teach, disclose, suggest, motivate, or render obvious applicants' invention as a whole. The rejection is in error and should thus be withdrawn.

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[3] In applicants' reading of the Little patent, it appears that the polypeptide of interest is first isolated, then immobilized on a surface, rather than being captured from inhomogeneous mixture on such probe.

CONCLUSION

Applicants respectfully submit that the claims are in good and proper form for allowance. If the Examiner believes, however, that there are any matters that remain outstanding before issuance, applicants respectfully request that the Examiner call the undersigned for a telephonic interview.

Respectfully submitted,

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